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Development and Physico-Chemical Analysis of *Varnya Lepa* Sunscreen Lotion (In-Vitro)

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ABSTRACT

Background: Direct exposure to Solar radiation causes many harmful effects on the skin like suntan, sunburn, hyperpigmentation and skin ageing etc. To overcome these types of problems sunscreen applications can be helpful. In Ayurveda, authors explained *Varnya Lepa* drugs like *Raktachandana* (*Pterocarpus santalinus* Linn.), *Manjistha* (Rubia cordifolia Linn), *Lodhra* (*Symplocos racemosa* Roxb), *Kustha* (*Saussurea lappa*), *Priyangu* (*Callicarpa macrophylla*), *Vatankura* (*Ficus benghalensis*) and *Masura* (*Lens culinaria* Medic) have a high potential due to their antioxidant activity, vitamin, photoprotective, flavonoids and phenolic acid in the safety of the skin from UV rays and blocks each UVA and UVB. This formulation is mentioned for external application in the form of *Lepa* but due to problems like stains after applications, impediment in utilization, protection trouble, non-accessibility of crisp medication all the season. If *Lepa* contains herbal drugs it should be used within 24 hours, otherwise, the drugs get decomposed and the application may harm the skin. Lotion is viscous semisolid preparation. They may be oil-in-water or water-in-oil type, easy to apply in unbroken skin without friction. Lotions contain water-soluble bases and hence are more convenient to use. **Aim:** The present study aim was to develop *Varnya Lepa* sunscreen lotion and Assessment of Sun Protection Factor (SPF) by In-vitro method. **Material and Methods:** A lotion base of Four trials was done, in this trial fourth was passed all parameters and the final development of sunscreen lotion was prepared. The quantity of extract used in sunscreen lotion was selected after assessment of the absorbance factor in UV-spectrophotometry and calculated sun protection factor. **Results**: The Sun protection factor was obtained 29 and shown under broad-spectrum sunscreen. **Conclusion:** *Varnya Lepa* sunscreen lotion was successfully developed. Hence, can be used as sunscreen lotion.

Key words: *UV radiation, Varnya Lepa Sunscreen Lotion, Sun Protection Factor.*

INTRODUCTION

Pores and skin is the outermost and largest part of the body. It's maximum sensitive to photodamage because it's mostly uncovered to UV radiation.[1] The harmful consequences of solar radiation are caused predominantly via the ultraviolet (UV) region of the electromagnetic spectrum, which may be divided into 3 regions: UVA, from 320 nm to 400nm; UVB, from 290 to 320 nm and UVC, from two hundred to 290 nm. UVC radiation is filtered out via the ecosystem earlier than attaining the earth. UVB radiation isn't completely filtered out by means of the ozone layer and is accountable for the damage because of sunburn and pyrimidine dimers. UVA radiation reaches the deeper layers of the epidermis and provokes the untimely getting old of the pores and skin, answerable for the generation of free radicals. UVB radiation is concerned in 65% harm of all skin.[2] Because of those dangerous results of UV radiation increases the chance of erythema of sunburn, suntan, skin most cancers and premature skin growing older and so forth. To save you those dangerous solar results, scientists formulated body lotions and creams where they added sunscreen like energetic substances

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to defend humans via absorbing, scattering or reflecting radiation.[3] The diverse artificial chemical sunscreens like aminobenzoic acid derivatives, anthranilates, benzophenones, cinematics, salicylates, inorganic sunscreens like titanium dioxide and zinc oxide can also cause mild to a moderate allergy in a few individuals. A number of those may additionally soak up into the bloodstream.[4] Natural formulations have more attractiveness due to their no facet effects and they're much less irritant.

Varnya-Lepa drugs like *Raktachandana* (*Pterocarpus santalinus* Linn.), *Manjistha* (*Rubia cordifolia* Linn), *Lodhra* (*Symplocos racemosa* Roxb), *Kustha* (*Saussurea lappa*), *Priyangu* (*Callicarpa macrophylla*), *Vatankura* (*Ficus benghalensis*) and *Masura* (*Lens culinaria* Medic) have antioxidant activity, vitamin, photoprotective, flavonoids and phenolic acid in the safety of the skin from UV rays and blocks each UVA and UVB. It has additionally properties consisting of wound restoration, antiseptic, anti-inflammatory, anti-cancer, anti-closma and promotes the complexion of the skin.[5] This *Varnya* formulation mentioned for external application in Lepa form. Due to usual problems with *Lepas* are stains after applications, impediment in utilization, non-accessibility of crisp medication in all the seasons. Herbal drugs containing *Lepas* should be used within 24 hours, otherwise the drugs get decomposed and the application may harm the skin. Lotion are viscous semisolid preparations. They may be oil-in-water or water-in-oil type, easy to apply in unbroken skin without friction. Lotions contain water soluble bases and hence are more convenient to use.[6] They'll be carried out without delay to the pores and skin with the help of some absorbent cloths which includes cotton wool or gauze soaked in it. The lotion used for local motion as cooling, soothing, or protective purposes.[7] Lotion containing suspended debris evaporates whilst done to the pores and pores and skin leaving a deposit of medicament on the ground. Lotions are less difficult to apply and less messy than many special outside arrangements.[8] A herbal lotion that could offer effective safety to the pores and skin and unfastened from any toxicity or poisonous residue or any infection at the same time as regularly used and must also be cosmetically proper. *Varnya Lepa* is one of the formulations which includes seven drugs like *Raktachandana* (*Pterocarpus santalinus* Linn.), *Manjistha* (*Rubia cordifolia* Linn), *Lodhra* (*Symplocos racemosa* Roxb), *Kustha* (*Saussurea lappa*), *Priyangu* (*Callicarpa macrophylla*), *Vatankura* (*Ficus benghalensis*) and *Masura* (*Lens culinaria* Medic). It is specifically indicated for melasma, enhances complexion and hyperpigmentation.[9] Olive oil used as one of the elements in the oil segment has antioxidant assets and still have UV filters.[10] However, it is necessary to develop *Varnya Lepa* sunscreen lotion and evaluate it sun protection factor in the cosmeceutical field to protect against solar radiation. The efficacy of sunscreen products has been identified as an vital public health trouble and it also includes expressed through the sun protection factor (SPF).[11] Determination of Sun Protector (SPF) changed into assessed by means of strategies In vivo and In Vitro method. In Vitro approach is a rapid, cost-effective reasonable device. It's far need of the hour to broaden more secure, chemically inert, nonirritating, nontoxic, photostable sunscreen merchandise, which can offer complete protection to the skin by means of the usage of novel natural formulations.[12]

The compound formulation *Varnya Lepa* was developed in the form of lotion and the Sun Protection Factor was evaluated by adopting the in-vitro method using UV spectrophotometer between the wavelength of 290 to 320 nm.

Aims and Objectives

1. To develop *Varnya Lepa Sunscreen Lotion*.
2. Assessment of Sun Protection Factor (SPF) by In vitro method.

Materials

Collection of raw drugs *Raktachandan, Manjista, Kustha, Lodhra* were procured from GMP certified KLE Ayurveda Pharmacy, Khasabag, Belagavi. *Priyangu, Vatankura* are collected from natural resources. *Masura*, Expecients and Preservatives are collected from Belgavi market.

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Identification and Authentication was done in AYUSH approved Drug Testing Laboratory for ASU drugs approved by Govt of India, KLEU's Shri B.M. Kankanwadi Ayurveda Mahavidyalaya, Shahapur, Belagavi, Karnataka.

METHODOLOGY

Preparation of *Varnya Lepa Churna*[13]

As per AFI All drugs i.e *Raktachandana, Manjistha, Kushta, Lodhra, Priyangu Masura and Vatankura* were taken 1 part each, grounded separately in pulverizer and passed through sieve size 40. The prepared *Varnya Lepa Churna* were stored in air tight plastic bags.

Hydroalcholic extraction of *Varnya Lepa Churna*[14]

Take 50gms of *Varnya Lepa* coarse powder in a conical flask. Add 1000ml of Ethanol and distilled water in the ratio 6:4, flasks were kept for 6 hrs continuous shaking and kept undisturbed for 18 hrs. Next day, extract was filtered through filter paper and filtrate is collected in beaker and put into Rota evaporator for removal of solvents till it becomes semisolid. This semi-solid extract is kept on Water Bath at 80°C to remove remaining moisture and prepared sticky consistency extract is stored in air tight container.

Development of *Varnya Lepa Sunscreen Lotion*[15]

The *Varnya Lepa Sunscreen Lotion* was developed as follows:

a) Pilot study to prepare lotion base

b) Final development of *Varnya Lepa Sunscreen Lotion*

A. The Pilot study were done in four Trials. In Trial Ist water phase and oil phase was mixed together and triturate till gets homogenous mixture and In Trial II to IV Trituration method was done by o/w method in Table no. 1.

Table 1: Pilot study of *Varnya Lepa* sunscreen lotion

SN	Ingredients	Trail I	Trial II	Trial III	Trial IV
1.	Olive oil	3ml	10ml	20ml	12ml
2.	Tween80	-	4gm	4gm	4gm
3.	Span80	-	1.5ml	3.5ml	3ml
4.	Glycerylmonostearate	1gm	2gm	2gm	2gm
5.	Sodium carboxymethyl cellulose	-	1gm	2gm	1.5gm
6.	Cetylalchol	0.5gm	-	-	-
7.	Glycerine	3ml	-	-	-
6.	Distilled water	42ml	40ml	40 ml	40 ml
7.	Quality Assessment	Not passed	Not passed	Not passed	Passed

Preliminary Quality assessment Parameters of lotion base after 24 hrs in Table No. 2

Table 2: Assessment parameters of *Varnya Lepa* sunscreen lotion

Trials	Creaming	Flocculation	Coalescence	Phase separation	Parameter
Trial-I	Negative	Negative	Positive	Negative	Not passed
Trial-II	Negative	Positive	Negative	Positive	Not passed
Trial-III	Negative	Positive	Negative	Positive	Not passed
Trial-IV	Negative	Negative	Negative	Negative	Passed

Observations of four trials in Preparation

1) **Trial I -** Emulsion base was formed soft, semisolid in nature, but get separated both phases after 24hrs (next day).

2) **Trial II -** Emulsion base was not formed, immediately get separated it was rejected.

3) **Trial III -** It was soft after gets separated after adding GMS.

4) **Trial IV -** Emulsion base was soft, semisolid consistency, easily spreadable, was suitable for lotion preparation.

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B) Final development of *Varnya Lepa Sunscreen lotion*

It was prepared with Trial IV ingredients quantity and extract ratio was selected after assessment of Sun Protection factor by UV-spectophotometric method in Table No. 3.

Table 3: Trial IV ingredients quantity and extract ratio.

Ingredients	VL 1	VL2	VL3	VL4	VL5	VL6
Varnya Lepa extract	0.5gm	1gm	2gm	2.5gm	3.5 gm	4gm
Olive oil	12ml	12ml	12ml	12ml	12ml	12ml
Tween80	4gm	4gm	4gm	4gm	4gm	4gm
Span80	3ml	3ml	3ml	3ml	3ml	3ml
Glycerylmonosterate	25ml	25ml	25ml	25ml	25ml	25ml
Carboxymethyl cellulose	1.5gm	1.5gm	1.5gm	1.5gm	1.5gm	1.5gm
Distilled water	53ml	53ml	53ml	53ml	53ml	53ml
Sodium Benzoate	0.5gm	0.5gm	0.5gm	0.5gm	0.5gm	0.5gm
Propylparabin	1.0gm	1.0gm	1.0gm	1.0gm	1.0gm	1.0gm
Methylparabin	0.05 gm	0.05 gm	0.05 gm	0.05 gm	0.05 gm	0.05 gm
Fragrance	Q.S	Q.S	Q.S	Q.S	Q.S	Q.S

In this Trial sixth was got highest Sun protection factor.

Table 4: Observation of *Varnya* Lepa Sunscreen Lotion.

Days	Trial VL I to VI
1st to 30th day	Prepared and preserved at room temperature
	Semiliquid consistency maintained
	Separation was not seen
	There is no change in colour and odour

Analysis of Sun Protection Factor (In-vitro)

The sun protection factor was analysed by UV-spectrophotometer and calculation was done.

Methodology: A sample application rate of 0.75mg/cm^2 is applied in small dots across the entire PMMA plate surface. The spreading motion for distributing the sample product should be light stokes applied in a shortest amount of time. Allow the sample plate to dry for 15min. as suggested in the operation manual of the UV-2000S Ultraviolet Transmittance Analyzer for the sample preparation and application technique. The sample thus prepared were exposed to Xenon flash lamp for determing the Sun Protection factor in wavelength of 290 to 400.

RESULTS

Table 5: The physico-chemical analysis of individual drugs

Drugs	Ash value	Acid insoluble ash	Water soluble extractive	Alcohol soluble extractive	Foreign matter	Loss on Drying
Raktachandana	1.841 %	0.248 %	2.636 %	4.392 %	Nil	6.364 %
Manjista	6.324 %	0.239 %	19.246%	5.210 %	Nil	5.527 %
Lodhra	11.343%	0.497 %	27.37 %	15.64 %	Nil	4.728 %
Kushta	2.685 %	0.781 %	20.299%	14.234%	Nil	6.427 %
Masura	2.398 %	0.449 %	29.520%	7.431 %	Nil	11.482%
Vatankura	1.690 %	0.397 %	2.955 %	2.271 %	Nil	11.597%
Priyangu	6.936 %	0.948 %	31.34 %	11.04 %	Nil	9.511 %

Table 6: Physico-chemical analysis of Developed lotion

SN	Tests	Results
1.	Form	Semisolids
2.	Colour	Reddish brown
3.	Odour	Pleasant

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4.	Touch	Smooth
5.	Appearance	Shiny
6.	Smoothness	Present
7.	Stickiness	Present
8.	Consistancy	Smooth
9.	Homogenecity	Present
10.	Spreadibility	Good
11.	pH (5% solution)	6.41
12.	Viscosity	1344cp

Table 7: *Varnya Lepa* sunscreen lotion Test for specified Micro-organism

Organism	Limits (As per IP)	Results
E-coli	Absent/100ml	Absent
S aureus	Absent/100ml	Absent
P eruginosa	Absent/100ml	Absent
S abony	Absent/100ml	Absent

Table 8: Microbial limit test (Quantitative)

Organism	Limits (As per IP)	Results
Total Bacterial count	30-300cfu/ml	21cfu/ml
Total Fungal count	10-100cfu/ml	08cfu/ml

Table 9: Determination of SPF analysis by In vitro[7]

Test Sample	Scans	1	2	3	Average values
Varnya Lepa Lotion	SPF	29.0	29.1	29.1	**29.0**
	Standard Deviation	0.18	0.17	0.18	**0.17**
	UVA/UVB ratio	0.897	0.895	0.896	**0.896**
	Critical wavelength	380.00	380.00	380.00	**380.00**

Images of *Varnya Lepa* sunscreen lotion Preparation

Fig. 1: *Varnya Lepa* extract

Fig. 2: *Varnya* extract + olive oil

Fig. 3: Mixing unidirectionaly

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Fig. 4: Trituration followed by Tween80 and Span80 till gets cracking sounds comes

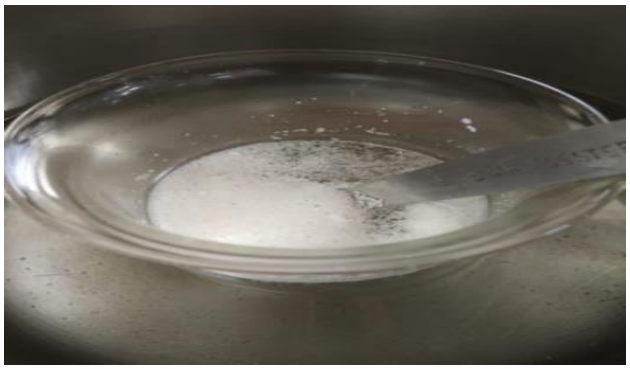

Fig. 5: GMS was dissolve in water heated upto 60°C

Fig. 6: GMS added into Lotion later CMC was added

Fig. 7: Trituration till gets homogenous mixed

Fig. 8: 50gms Packaging

DISCUSSION

All the *Varnya raw drugs* are complies the API standards and taken for the preparation. While preparing *Varnya Lepa Churna* it is observed the maximum loss in *Raktachandana, Kushta* and *Lodhra* due to its hard nature. The ratio of ethanol and Distilled water was selected 6:4 only after alcohol and water soluble extractive of individual raw drugs was done. It was indicated that *Varnya* drugs are having more ethanol soluble than water soluble. Rota evaporator was used to remove aqueous part by this instrument we can preserve the phytoconstituents. *Varnya* extract was not fully converted in to powder form due to the presence of tannin and starch get bounded to form sticky material.

In Pilot study, cetylalcohol was used in Ist Trial which was immediately separated. From Trial II to IV cetylalcohol was replaced with Tween80, Span80 which helps to create good emulsion and also acts as good solubilizer.[16] Sodium carboxymethyl cellulose having more capacity to absorb or hold water acts as binding agent.[17] The quantity was selected accordingly. Olive oil contains three major antioxidants i.e., Vit-E, polyphenols and phyto sterols that are responsible for radio protective activity.[18] Most of *Varnya* Drugs are shown presence of Flavanoids. Studies have shown that Flavonoids have a lot more antioxidant activity than Vit C and E which helps to protect skin from UV radiations and improves skin texture.[19]

Manjista, Kushta, Lodhra, Vatankura, Masura and *Priyangu* are having presence of Saponins, Glycosides protects against UV damage by inhibiting extracellular matrix degradation, Skin tonifying, reduces erythema and having antiaging property.[20] The pH of *Varnya Lepa sunscreen lotion* was 6.4, so it is not harmful for the skin and also under the normal limit of local applications. Spreadibility was good, easily spreads. Viscosity was viscous in nature and after application covers all the skin surface, good consistency. Microbial Limit Test was Under the IP limits. The developed lotion Sun Protection Factor was shown value 29. The good sunscreen products available in the market claimed the

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SPF value ranging between 15-30. By adding herbal ingredients in *Varnya Lepa Sunscreen lotion* was also be claimed under broad spectrum Sunscreen lotion.

CONCLUSION

It is concluded that *Varnya Lepa Sunscreen lotion* was successfully developed. Sun protection Factor (SPF) of *Varnya Lepa Sunscreen lotion* was 29.0 which shown under broad spectrum Sunscreen lotion. Hence, it can be used as Sunscreen lotion.

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